Available online on 15.05.2019 at http://jddtonline.info

Journal of Drug Delivery and Therapeutics

Open Access to Pharmaceutical and Medical Research

Open Access Research Article

A New simple and sensitive method for simultaneous estimation of Aspirin and Omeprazole in rat plasma by RP-HPLC and its application to pharmacokinetic study

Patta Salomi[1*], M. Purushothaman[2], S.V. Satyanarayana[3]

[1] Research Scholar in JNTUA, Ananthapuramu-515002, India

[2] Scient Institute of Pharmacy, Ranga Reddy District -501506, India

[3] Department of Chemical Engineering, JNTUCEA, JNTUA, Ananthapuramu -515002, India

ABSTRACT

A new reverse phase HPLC method for simultaneous estimation of Aspirin and Omeprazole in rat plasma was developed and validated. The separation was achieved on Hypersil ODS C18 column. The mobile phase Acetonitrile: Methanol: 0.05M Phosphate buffer (40:5:55) was delivered at a flow rate of 1ml/min. The Eluent was monitored using spectrophotometric detection at 225nm. The relationship between Aspirin and Omeprazole and peak height ratio was linear over the range of 5-15µg/ml and 3-8.7µg/ml respectively. Percentage recovery for Aspirin was 100.02 and Omeprazole was 99.66. The Tmax for Aspirin was 1hr 20mins and for Omeprazole was 1hr 15mins respectively. Prepared Aspirin and Omeprazole levels after oral administration of a therapeutic dosage were 3.1ug/ml and 1.8µg/ml indicating that the described method is potentially suitable for TDM.

Keywords: Aspirin, Omeprazole, rat plasma, Protein Precipitation, RP-HPLC.

Article Info: Received 30 March 2019; Review Completed 06 May 2019; Accepted 10 May 2019; Available online 15 May 2019

Cite this article as:

Salomi P, Purushothaman M, Satyanarayana SV, A New simple and sensitive method for simultaneous estimation of Aspirin and Omeprazole in rat plasma by RP-HPLC and its application to pharmacokinetic study, Journal of Drug Delivery and Therapeutics. 2019; 9(3):332-338 **http://dx.doi.org/10.22270/jddt.v9i3.2673**

***Address for Correspondence:**

Patta Salomi, Research Scholar in JNTUA, Ananthapuramu-515002, India

Introduction

Yosprala is an only sequentially designed therapy in combination of aspirin and omeprazole for preventing gastric ulcers produced by aspirin used to prevent cardio and cerebrovascular events. Aspirin is a salicylate which acts as on the substances by reducing pain, fever and inflammation. Omeprazole is a proton pump inhibitor which acts by decreasing the production of acid secretion in the stomach. Yosprala is a platelet aggregation inhibitor. Thus, it is used to lower the various cardiac problems such as strokes or death.

In certain conditions the problems are related to blood clots, angina, past heart attack and stroke or mini strokes. To improve the blood flow to the heart for the patients who had surgery are treated with yosprala. Omeprazole in this combination acts to reduce the gastric ulcers produced by aspirin. The individual dosage of aspirin and omeprazole will not have same therapeutic activity as of combination as yosprala. [1] Yosprala [fig-1] is a combination of salicylate and proton pump inhibitor, chemically it is called as yosprala; aspirin/omeprazole; omeprazole/aspirin; aspirin with the mixture of yosprala. IUPAC name of yosprala is 2acetyloxy benzoic acid; 6-methoxy-2-[(4-methoxy-3, 5-dimethyl pyridine-2-yl methyl sulfinyl]-H-benzimidazole. Its molecular weight is 525.576g/mol, corresponding to the molecular formula $C_{26}H_{27}N_3O_7S$. It is freely soluble in methanol and water and partially soluble in ethanol and acids. [2]

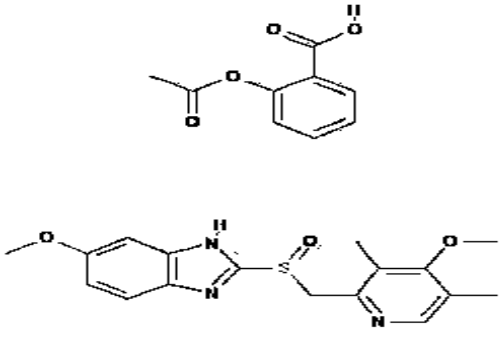

Fig-1 . Structure of Yosprala

Yosprala is a delayed release tablet orally administered.One tablet of yosprala[fig-2] contains 1 strength of omeprazole and 2 strengths of aspirin in two different strengths as 81/40, 325/40.It should be administered 1hour before meal and should be swallowed whole with liquid.It is mostly indicated for patients who require aspirin for secondary prevention of cardiovascular and cerebrovascular events and who are at high risk of producing gastric ulcers due to daily dosage of aspirin.It is highly caused in people above 55years of age and or patients having past ulcerative problems.[3]

Mechanically as it is a combination drug aspirin acts as antiplatelet agent which inhibits the synthesis of prostaglandin and aggregation of platelets and omeprazole is a substituted benzimidazole which belongs to the class of antisecretory compounds.It shows its activity by suppresing the release og gastric acid by inhibiting the [H+/K+]ATPase enzyme which is responsible for production of gastric acid on the surface by parietal cells.Yosprala may cause side effects such as gastritis,diaarhoes,gastric polyps,nausea and non cardiac chest pain.[4]

Because of it's therapeutically importance quantitative determination of yosprala is very significant in pharmaceuticals and human physiological fluids for both quality control of preparations and chemical diagnosis. In the last years many methods have been reported for the determination of aspirin and omeprazole individually and very few attempts have been developed for estimation of yosprala. The current reviews surveys the methods developed to determine yosprala in drug products.

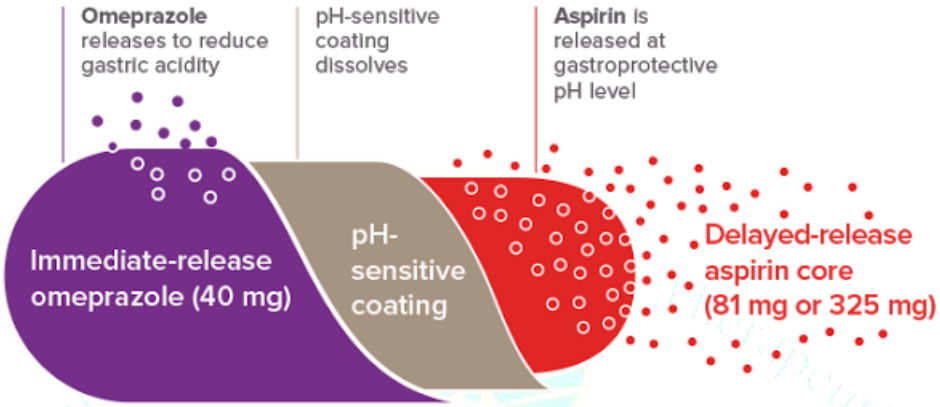

Fig-2. Yosprala capsule

Methods for pharmaceuticals

Chromatographic Methods

Performance characteristics of RP-HPLC Method

S.No	Stationary phase, Mobile Phase	UV-detection (nm)	Linear range (µg/ml)	LOD	LOQ	Application	Ref
1	C_{18} (150mm-3.0 mm,5 µm); 0.1M Dipotassium phosphate buffer (pH 3) and Methanol in the ratio of 40:60v/v	246nm	10-50µg/ml for Aspirin and 50-250µg/ml for Omeprazole	9.24 µg/ml	10.37 µg/ml	Bulk and Dosage form	[5]
2	C_8 (150×4.6, 5um) column quaternary gradient pump SP930D HPLC system. Acetonitrile:Methanol:Phosphate Buffer at pH 2.5 in the ratio 25:10:65	228nm	20-100µg/ml for -AP10-50 µg /ml for -OMP	1.23 µg/ml –ASP 1.25 µg/ml-OMP	4.12 µg/ml – ASP, 4.18 µg/ml – OMP	Dosage	[6]
3	C_{18} Ammonium Acetate Buffer+Acetonitrile+Methanol	233nm	18-42µg/ml for -ASP 6-14µg /ml for -OMP	2.880 µg/ml –ASP 1.987-µg/ml – OMP.	6.703 µg/ml – ASP 4.630 µg/ml - OMP.	Tablet dosage form	[7]
4	C18 column,Acetonitriel-water (60:40v/v)	240nm	10-100 µg/ml for –ASP 4-80 µg/ml for -OMP			Bulk powder and pharmaceutical formulation	[8]
5	Agilent C18 column, buffer Ph	240nm	10 µg/ml for –ASP 10 µg/ml for -	0.1372	0.132-ASP	Capsule dosage form	[9]

	3:Methanol(30:70)		OMP	-ASP 3.17-OMP	5.60-OMP		
6	Zorbax Eclipse XDB-C18;Acetonitrile:water(50:50,v/v)	293nm	8.1 µg/ml for –ASP 4 µg/ml for -OMP	0.67 µg/ml for –ASP 0.43 µg/ml for -OMP	1.94 µg/ml for –ASP 1.25 µg/ml for -OMP	Bulk sample powder	[10]
7	C18 water xterra;water;Methanol(50:50v/v)	244nm	81 µg/ml for –ASP 40 µg/ml for -OMP	0.04517 µg/ml for –ASP 0.003620 µg/ml for -OMP	0.1369 µg/ml for –ASP 0.01097 µg/ml for -OMP	Synthetic mixture	[11]
8	Thermo C18 column;Acetonitrile:phosphate buffer(70:30v/v)	295nm	5-25 µg/ml for –ASP 5-25 µg/ml for -OMP	0.89 µg/ml for –ASP 0.45 µg/ml for -OMP	2.41 µg/ml for –ASP 1.25 µg/ml for -OMP	Bulk drug and Synthetic mixture	[12]
9	Zodiac C18 column;0.1Mpotassium dihydrogen phosphate: Acetonitrile (55:45%v/v)	260nm	0.65-195 µg/ml for –ASP 0.08-24 µg/ml for -OMP	0.199 µg/ml for –ASP 0.024 µg/ml for -OMP	0.650 µg/ml for –ASP 0.080 µg/ml for -OMP	Tablet dosage form	[13]
10	Cosmosil C18column;Methanol:Water (60:40)	231nm	10-50 µg/ml for –ASP 20-100 µg/ml for-OMP	0.5375 µg/ml for-ASP 0.5946 µg/ml for-OMP	1.6290 µg/ml for-ASP 1.9820 µg/ml for-OMP	Bulk and Dosage form	[14]

UVvisible spectrophotometric method

S.No.	Diluent	λmax (nm)	Linear range(mg/mL); molarextinction coefficient	LOD	LOQ	Application	Ref
1	Methanol	224.7nm- ASP 251nm-OMP	0.5-25µg/ml –ASP 1-8µg/ml -OMP	1.08 µg/ml	3.27 µg/ml	Bulk dosage form	[15]
2		274nm-ASP 302nm-OMP	25-125 µg/ml for- ASP 3-15 µg/ml for-OMP			Laboratory Sample	[16]

The review of the analytical methods reported for Aspirin and Omeprazole [YOSPRALA] showed that UV-spectrophotometric and RP-HPLC methods were developed and validated by using different solvents. The reported methods [17-23] are developed to determine Aspirin individually or in combination with other drugs and the methods [24-30] are developed to determine Omeprazole individually or in combination with other drugs using different techniques. No bio analytical method yet has been reported. Hence an attempt is made to develop a method for simultaneous estimation of Aspirin and Omeprazole in rat plasma and the proposed method was applied for pharmacokinetic study.

Experimental

Apparatus

Chromatography was performed on Waters HPLC that includes photodiode array detector and an auto injector. A reverse phase Thermo scientific Hypersil ODS(250x4.6mm,5mm) Column in conjugation with guard packed pre-column and C18 insert were used for separation.

Chemicals and Reagents

All reagents were of analytical grade unless stated otherwise. Acetonitrile HPLC grade was supplied by Yarrow Chem Products Mumbai. Water for HPLC analysis were generated by RO, UV, MilliQ water. Aspirin,(CAS N0:50-78-2),Potency:99% , Expiry:2/2020 Omeprazole (CAS NO:73590-58-6),Potency: 99% ,Expiry: 2/2020.

Chromatographic Conditions

The mobile phase consisted of 0.05M Phosphate buffer (Ph 4 adjusted with 0.1% Tri ethyl Amine),Methanol and Acetonitrile in (55:5:40). Before delivering in to the system, the mobile phase was filtered through 0.45µm polytetrafluroethylene filter and Sonicated for 5min. The analysis was carried out under isocratic conditions using flow rate of 1ml/min at room temperature [260c]. Chromatograms were recorded at 225nm using photodiode array detector.

Stock Solutions

Stock solutions of aspirin and omeprazole were prepared by dissolving 10mg of aspirin and 5mg of omeprazole in 100ml of 0.05M phosphate buffer to enhance solubility. The solution was shaken completely by hand and Sonicated for 10minutes. The concentration of both stock solutions were equivalent to 1000µg/ml concentrate of aspirin and 500 µg/ml concentration of omeprazole [STOCK SOLUTION –A]

1ml of stock solution of aspirin and omeprazole were added to 10ml of blank plasma to produce a solution with concentration of 100 µg/ml of aspirin and 50µg/ml of Omeprazole.[STOCK SOLUTION-B]

Working Standards

1ml of stock-B is added to 10ml of mobile phase to produce 10 µg/ml concentration of Aspirin and 5µg/ml concentration of omeprazole. The solutions were vertexed for 1min and 0.5ml aliquot was transferred to 1.5ml micro centrifuge tubes and stored at 20^{o}c until used.

Calibration Standard /Q.C. Samples

Calibration standards were prepared by mixing nine volumes of aspirin and omeprazole working solutions in the range of 2-40 µg/ml concentration for aspirin and 1-20 µg/ml for Omeprazole. Six quality control samples were prepared mixing 0.5,1,2,3,4 and 5ml of stock-B solutions to 10ml of blank rat plasma to yield concentrations of 5, 10,20,30,40 and 50 µg/ml concentrations of aspirin and 2.5,5,10,15,20 and 25 µg/ml concentrations of Omeprazole. The solutions were vertexed for 1ml then 0.5ml of aliquots were transferred to 1.5ml Eppendrof micro centrifuge tubes and stored at -200c until used.

Sample Preparation

0.5ml of quality control samples were placed in 1.5ml micro centrifuge tubes. The solutions were vertexed for 30sec, Sonicated for 1min, transferred centrifree Micro partition system, precipitated using methanol and centrifuged at 3000rpm for 30min.0.1ml of ultra filtrate was injected in to chromatographic system using an auto sampler.

Pharmacokinetic Study

Animal

A male albino rat(200g)housed under standard conditions and had a labium access to water and standard laboratory diet throughout the experiments were used in the present study. After a single dose by oral administration of 15mg/kg and 8mg/kg of Aspirin and Omeprazole, Blood samples were collected at regular intervals of time up to 24hrs.

Preparation of ASP and OME oral doses

ASP and OME was dissolved in water by using 1%Tween 80 and 0.5% carboxyl methyl cellulose.ASP and OME were given to wistar rats with oral dose of 15mg/kg and 8mg/kg respectively.

The developed IL-DLLME method was successfully used to quantification of Aspirin and Omeprazole concentration after 15mg/kg oral administration of Aspirin and Omeprazole in six rats. The standard calibration curve was used to determine the sample concentrations in the unknown samples.

Results and discussions

Aspirin and Omeprazole were resolved with resolution factor greater than 3.2 with a routine of 8minutes. To evaluate assay specificity placebo run and blank run was performed and none was found to calculate with Aspirin or Omeprazole. The plasma calibration curves were constructed using peak height ratios of Aspirin and Omeprazole and their concentrations. Linear regression analysis was used to calculate slope, intercept and co relation co efficient (r^2). The linearity over the range of 5-15 µg/ml for Aspirin at 3-8.7 µg/ml for Omeprazole was found to be satisfactory and reproducible over the time, r^2 obtained was 0.9994 for Aspirin and 0.9998 for Omeprazole.

Accuracy of assay determined at Aspirin and Omeprazole concentration of over three different days. Accuracy was in the range 99.7-102 percent. Relative standard deviation for precision data obtained for was 0.23 and 0.34 for Aspirin and Omeprazole respectively. The extraction recovery for Aspirin and Omeprazole ranged from 99.8 to 100.2 and the result is represented in Table-1 &2

Table-1 Accuracy and Precision data for Aspirin

S.N0	Nominal concentration(µg/ml)	Concentration found(µg/ml)	Peak area	%recovery
1	5	5.11	25236	102±0.02
2	5 (50%)	5.03	25140	100±0.06
3	5	5.04	25141	100±0.08
4	10	10.04	51329	100±0.04
5	10 (100%)	10.03	51328	100±0.03
6	10	10.08	51335	100±0.08
7	15	15.01	75423	100±0.01
8	15 (150%)	15.12	75502	100±0.08
9	15	15.05	75431	100±0.03
		Standard Deviation S.D.=1.36		**Relative standard deviation R.S.D=0.135**

Table-2 Accuracy and Precision data for Omeprazole

S.N0	Nominal concentration(µg/ml)	Concentration found(µg/ml)	Peak areas	%recovery
1	3	3.01	12638	100±0.001
2	3	3.00	12637	100±0.000
3	3	3.02	12639	100±0.002
4	5.9	5.88	24966	99±0.66
5	5.9	5.91	24970	100±0.001
6	5.9	5.92	24971	100±0.002
7	8.7	8.73	36795	100±0.034
8	8.7	8.71	36786	100±0.001
9	8.7	8.72	36787	100±0.002
		Standard Deviation S.D.=0.776		**Relative standard deviation R.S.D=0.132**

In regard to system suitability parameters ruggedness and robustness and significant effect was observed using two different HPLC instruments. Further the method was found to be reproducible from one analyst to another.

Fig-3, 4 & 5 shows chromatograms of Aspirin and Omeprazole standards; 10, 20, 30 µg/ml concentration of Aspirin and 5, 10, 15 µg/ml concentration of Omeprazole.

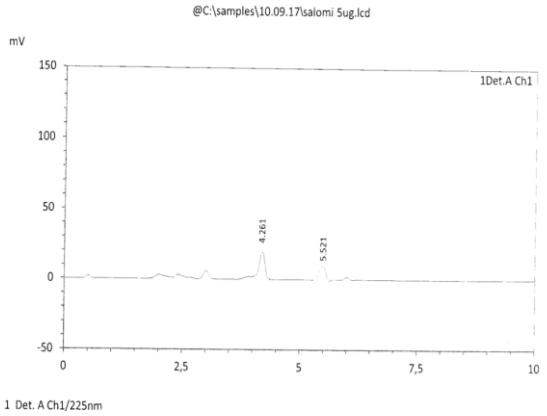

Fig-3 – Chromatogram of Aspirin and Omeprazole (5ug/ml)

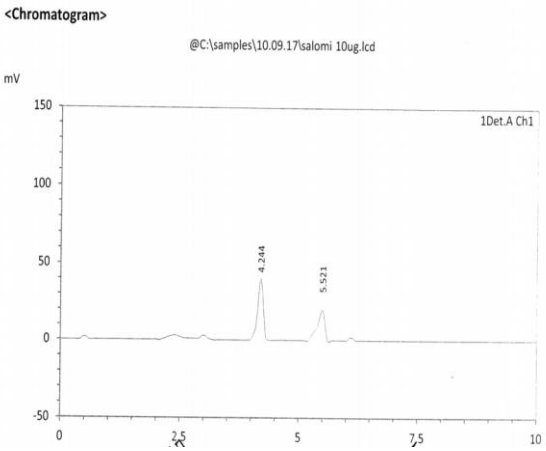

Fig-4 – Chromatogram of Aspirin and Omeprazole (10ug/ml)

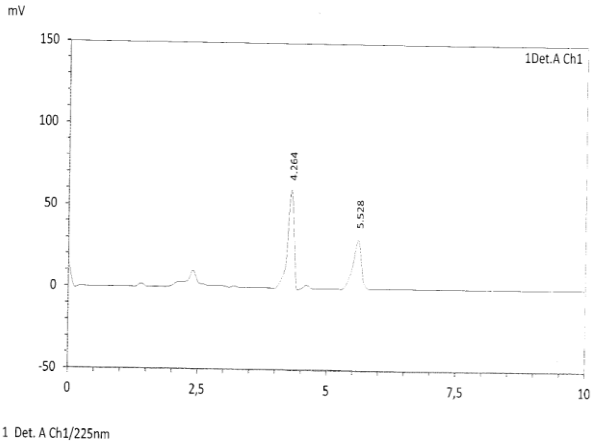

Fig-5– Chromatogram of Aspirin and Omeprazole (15ug/ml)

TABLE-3 represents the peak areas obtained for Aspirin and Omeprazole and the concentrations obtained for blood sample withdrawls from rats.

Table-3- Peak areas and concentrations of drugs

Code	Decode	Aspirin		Omeprazole	
		AUC	Conc. ug/ml	AUC	Conc. Ug/ml
5	5 ug	25134	5.0	12637	3.0
10	10 ug	51326	10.2	24968	5.9
15	15 ug	75421	15.0	36785	8.7
a	0.5h	842	0.2	637	0.2
b	1h	13256	2.6	6842	1.6
c	1.5h	14695	2.9	6891	1.6
d	2h	5802	1.2	2865	0.7
e	2.5h	862	0.2	567	0.1

- **Code 5, 10, 15- Concentrations of aspirin prepared.**
- **a,b,c,d,e- Blood sample withdrawals at timely basis**

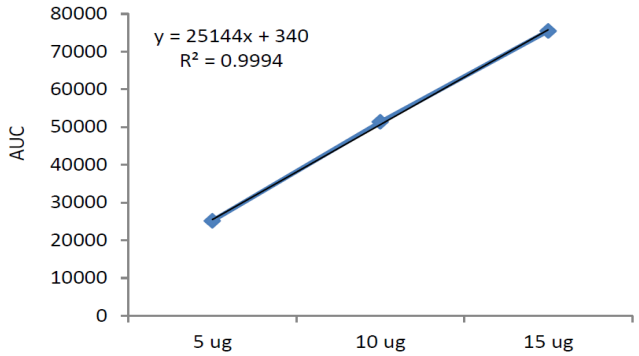

Fig-6-Calibration curve for various concentrations of Aspirin

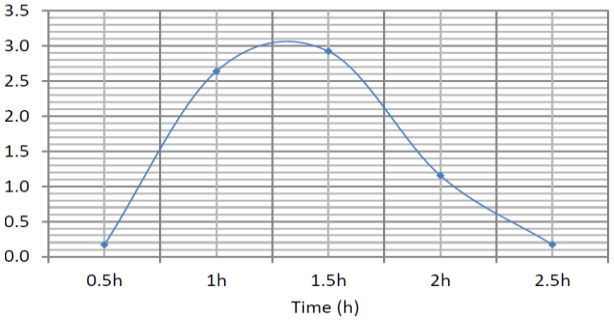

Fig-7- Time vs concentration curve for Aspirin

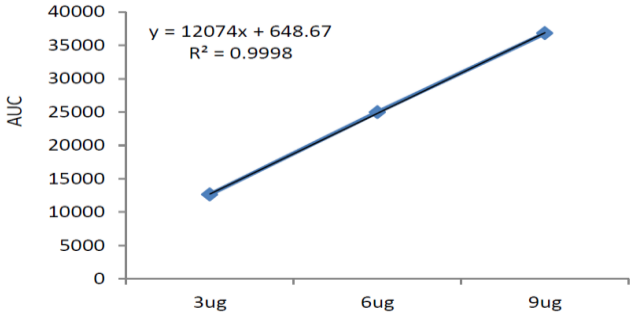

Fig-8-Calibration curve for various concentrations of Omeprazole

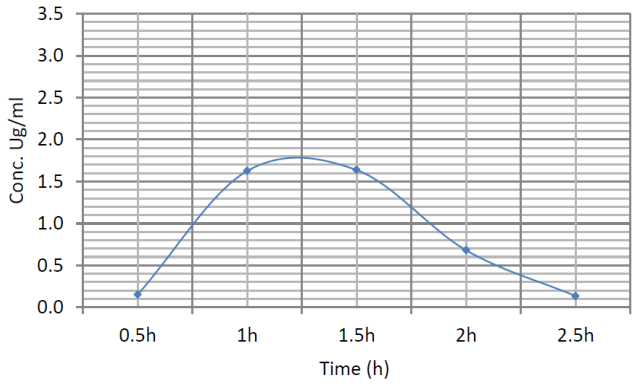

Fig-9-Time vs. concentration curve for Omeprazole

Conclusion

This is the first report submitted which includes method development to determine Aspirin and Omeprazole in rat plasma and application of method for pharmacokinetic study. In summary, the Describe method is rapid, sensitive, specific, accurate and reproducible. It is of potential value for determination of Aspirin and Omeprazole in plasma by RP-HPLC in therapeutic levels. The Tmax for Aspirin was 1hr 20mins and for Omeprazole was 1hr 15mins respectively. Prepared Aspirin and Omeprazole levels after oral administration of a therapeutic dosage of tablet were 3.1ug/ml and 1.8µg/ml indicating that the described method is potentially suitable for TDM.

Acknowledgement

We are thankful to P.Rami Reddy Memorial College of Pharmacy for providing facilities to carry out the research work.

Conflicts of Interest

No conflicts of interest

References

1. https://aralez.com/portfolio/yosprala/

2. https://googleweblight.com/i?u=https://www.drugs.com/yosprala.html&grqid=EV6RH6dO&hl=en-1N

3. https://googleweblight.com/i?u=https://www.yospralahcp.com/&grqid=F1TNO&hl=en-1N

4. Center watch for drugs –yosprala

5. Vani R, Sunitha M, analytical method development and validation for the determination of omeprazole and aspirin using RP-HPLC method in bulk and dosage form, 2017; 2(4).

6. Kalshetti MS, Kasabe S, Chauhan N, Dhanshri S, Kale B, Development and validation of RP-HPLC method for simultaneous estimation of aspirin and omeprazole in dosage. (2017); 2(4):45-51.

7. Kranthi KK, Supriya D, Divya D, Rani D, Neelima Munn G, Analytical method development and validation for the estimation of aspirin and omeprazole using RP-HPLC method. (2017); 4(1):044-071.

8. Mohammed W.I.Nassar, Khalid A.M.Attia, Ahmed A.Mohamed, Mohamed Shahin, HPLC method for simultaneous estimation of Aspirin and Omeprazole in their new combination, journal of Analytical chemistry Letters. 2017; 7(3):438-444.

9. Bonagani N, Manda RM, Nikitha V, Prashanth G, Santhosh N, Praveen Reddy D, A Validated RP-HPLC method for aspirin and omeprazole in its pure and capsule dosage form, International Journal of Pharmaceutical Biological and Chemical Sciences. 2017; 6(4):1-14.

10. Khan HN, Bandewar SS, Zameeruddin M, Bharkad VB, Development and Validation of RP-HPLC Method for simultaneous determination of Aspirin and Omeprazole, Der pharma Chemica, 2017; 9(20):55-58.

11. Gawande VV, Chandewar AV, A validated HPLC-DAD method for combined aspirin and omeprazole synthetic mixture, Scholars Research Library, 2018; 10(4):57-67.

12. Jain AK, Dubey BK, Basedia D, Khare S, Dhakar S, Om Prakash, New cost effective stability indicating RP-HPLC method for simultaneous determination of Aspirin and Omeprazole in the bulk drug and synthetic mixture, Asian journal of Pharmaceutical Education and Research, 2018; 7(3):105-115.

13. Yenduri G, Navuluri S, Analytical HPLC method for estimating the combination of Aspirin and Omeprazole in bulk and tablet dosage form, MARMARA pharmaceutical journal, 2018; 22(4):502-510.

14. Sarode T K, Jadhav PB, R-HPLC method development and validation for simultaneous estimation of Aspirin and Omeprazole bulk and dosage form. 2018; 8(5):322-328

15. Salomi P, Sultana A, Sharmila T, Nagarajan G, Gnana Prakash K, Simultaneous estimation of aspirin and omeprazole (yosprala)in bulk by UV-spectroscopy 2017; 3:87-91.

16. Rakesh, Simultaneous estimation of aspirin and omeprazole in laboratory sample by different Uv spectrophotometric Techniques, Indian Drugs, 2018.

17. Suresh Kumar S, Jamadar LD, Bhat K, Musmade PB, Vasantharaju SG, Udupa N, Analytical method development and validation for Aspirin, J. International Journal of ChemTechResearch, 2010; 2(1):389-399.

18. Sanjaypai PN, Gaude S, Simultaneous Estimation of A Three Component Mixture of Aspirin, Caffeine and Orphenadrine Citrate by UV Spectrophotometric Method of Absorbance Correction for Interference, J. Indian journal of applied research, 2016); 6(2):717-719.

19. Khristi AP, Mardia RB, Suhagia BN, UV Spectrophotometric method development and validation of first derivative method for simultaneous estimation of sildenafil citrate and aspirin in bulk and tablet dosage form, J. Indo American Journal of Pharmaceutical Research, 2015; 2837-2843.

20. Murtaza G, Khan GA, Shabbir A, Mahmood A, Asad MHHB, Farzana K, Malik NS, Hussain I, Development of a UV-spectrophotometric method for the simultaneous determination of Aspirin and Paracetamol in tablets. Scientific Research and Essays, 2011; 6(2):417-421

21. Bhiji PMC, Pandya C, Development and validation of RP-HPLC and UV method for simultaneous quantitation of Clopidogrel Bisulphate and Aspirin bulk drug, J. Trade science analytical chemistry and Indian journal, 2014; 15(2):43-48.

22. Chodvadiya FJ, Thula KC, Maheshwari DG, Simultaneous estimation of aspirin and lansoprazole by rp–hplc method, J. International Journal of Recent Scientific Research, 2015; 6(4):3385-3390

23. Patel B, Parmar S, Doshi J, Captain AD, Development and Validation of HPTLC Method for Simultaneous Estimation of Esomeprazole Magnesium and Aspirin in Bulk and Synthetic Mixture, J .International Journal for Pharmaceutical Research Scholars (IJPRS), 2014; 3(1):2277-7873.

24. Kumaraswamy D, Rathinaraj PS, Rajveer CH, Sudharshini S, Shrestha B, Rao PR, Process validation of analytical method development and validation for Omeprazole capsules and blend, J. International Journal of Pharma and Bio Sciences, 2010; 1(2):1-6.

25. Jadhav S, Kharat R, Pirjade MF, Tamboli A, Zero order and area under curve spectrophotometric methods for determination of Omeprazole capsules in pharmaceutical formulation, J. International Journal of Advances in Scientific Research, 2015; 1(2):102-107.

26. Vijayaraghavan R, Jayababu G, Prasad R, Thirugnanam PE, Gayathri S, Sriraam VT, Ramesh Kumar G, Bio-analytical method development and validation for Omeprazole using LC-MS/MS, J. International journal of pharmaceutical sciences and research 2011; 2(9):2475-2481.

27. Kalakonda SN, Mohammad BD, KalyaniP, DussaKK, Development and validation of RP-HPLC method for the estimation of Omeprazole in bulk and capsule dosage forms, J. International Current Pharmaceutical Journal, 2012; 1(11):195-205.

28. Nagarajan G, Nagesh P, Ramana BV, Ratna Prasanna N, Triveni C, Development and validation of RP-HPLC method for simultaneous estimation of Omeprazole and Cinitapride in bulk and capsule dosage form, J .International Journal Of Pharmacy, 2013; 4(2):131-135.

29. Kulkarni AS, Mane VB, Method development and validation for the simultaneous determination of Omeprazole and Domperidone in solid dosage form by RP-HPLC, J. International Journal of Pharmacy and Pharmaceutical sciences 2012; 4(5):109-114

30. Topagi KS, Jeswani RM, Sinha PK, Damle MC, A validated normal phase HPLC method for simultaneous determination of Drotaverine hydrochloride and Omeprazole in pharmaceutical formulation, J. Asian Journal Of Pharmaceutical and Chemical Research, 2010; 3(1):1118-1121